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IMMUNOCHEMICAL INVESTIGATIONS OF CELL SURFACE ANTIGENS

OF NEISSERIA MENINGITIDIS AND NEISSERIA CONORRHEA

Final Report

April 1979

Ъу

Frederic A. Wyle, M. D.

Supported by

US Army Medical Research and Development Command Fort Detrick, Frederick, Maryland 21701

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University of California School of Medicine Irvine, California 92664

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Cell surface antigens of N. gonorhoeae and N. meningitidis were isolated and purified. Antisera to these antigens were produced in rabbits and used to examine their species specificity. The cell mediated immune response in patients with clinical gonococcal infections were examined using these anti-		

gens. It was found that such patients have leukocyte transformation responses to both gonococcal and meningiococcal antigens. Responses in female patients

were found to be much greater than that of male patients

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Final Report

Contracting Officer HQDA (SGRD-MD)
Research and Development Command Washington, D. C. 20314

Dear Sir:

The work outlined in this final report falls into three categories: (1) isolation, purification and chemical analysis of cell surface antigens of Neisseria gonorrhoeae and Neisseria meningitidis; (2) determination of the serological specificity of the cell surface antigens of N. gonorrhoeae and N. meningitidis; and (3) investigations into the cellular immune response of patients with uncomplicated gonorrhea to purified cell surface antigens of N. gonorrhoeae and N. meningitidis.

Isolation of N. gonorrhoeae and N. meningitidis cell surface antigens was accomplished via a modification of the Ribi ether extraction technique. Partial purification of crude antigens was effected by ribonuclease and deoxyribonuclease treatment together with gel filtration chromatography. This procedure resulted in a two peak elution pattern which was virtually identical for the gonococcal and meningococcal antigens. Chemical analysis showed composition of the first peak to be primarily protein, while the second peak was largely nucleic acid. These purification steps were performed on numerous gonococcal and meningococcal strains stocked in the laboratory. Comparison of the antigenic structure of the outer membrane for these strains was carried out utilizing sodium dodecyl sulfate polyarylamide gel electrophoresis.

The antigens designated above were utilized to produce antisera in rabbits. The antisera were tested by immunodiffusion, countercurrent immunoelectrophoresis, and indirect hemagglutination techniques. These serological studies revealed extensive cross-reactivity between crude gonococcal and meningococcal antigenic preparations. Partial purification reduced the degree of cross-reactivity detected by the tests.

Peripheral blood lymphocytes (PBL) were isolated from blood samples obtained from patients with uncomplicated gonorrhea at either the Santa Ana Venereal Disease Clinic or the University of Southern California-Los Angeles County Medical Center Venereal Disease Clinic. PBL transformation stimulated by gonococcal and meningococcal antigens were utilized as a measure of cell-mediated immunity (CMI). Both male and female patients exhibited a broad range of blastogenic responses to gonococcal and meningococcal antigens. Control subjects demonstrated significantly lower and more uniform responses.

As a group, female patients displayed a greater CMI response than male patients. Cross-reactivity between N. gonorrhoeae and N. meningitidis was manifested by PBL transformation in patients with gonorrhea to non-purified menigococcal antigen. Partial purification of the meningococcal antigen by gel chromatography resulted in a reduced CMI response in both male and female patients. Female patients demonstrated marked stimulation with the purified gonococcal antigen, while male patients showed only slight stimulation. This study verified the lymphocyte transformation response in gonococcal infection. The functional role of CMI in gonococcal infection remains uncertain.

Sincerely,

FREDERIC A. WYLE, M.D. Assistant Professor of Medicine

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